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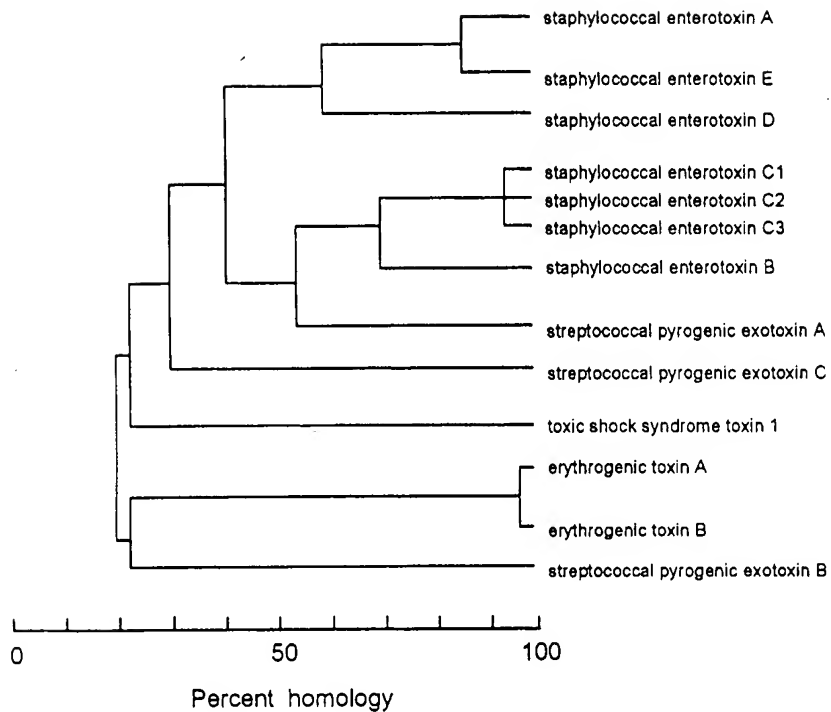


Fig 1

FIGURE 1

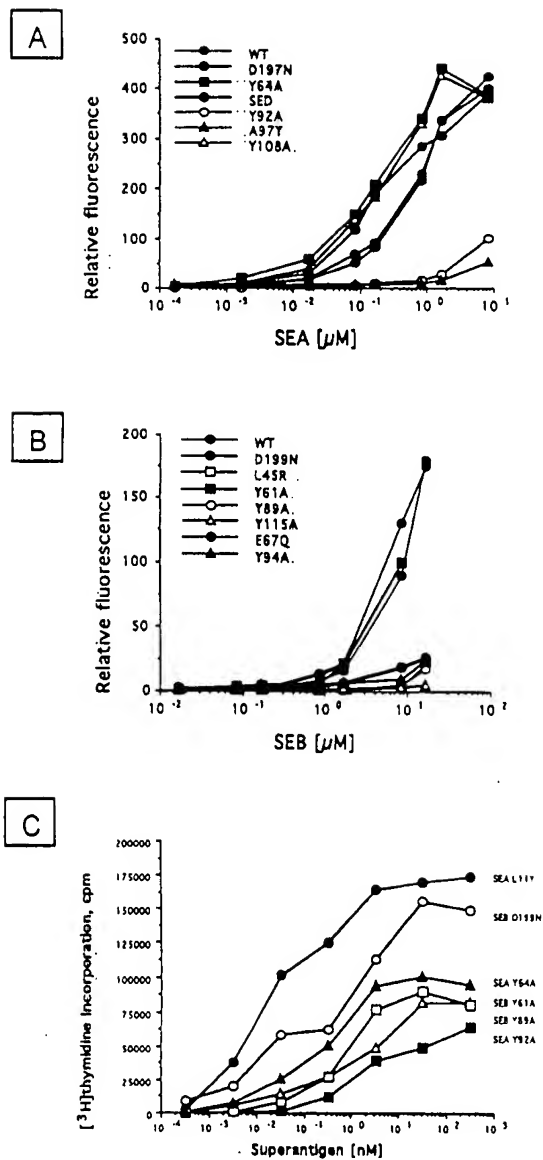


Fig. 2

SEA	...SHDQF	QHTILFKGFFTDH	SWNDLLV	FDSKDIVDKYK	GKKVDLYGAY	GYQCA	.....GGTPNK	TACM	GGVTLHDNNRLTEKK
SED	...TGDOF	ENTLLYKFFFTDL	INFEDLLI	FNSKEMAOHFK	SKNVDVYPIR	SINCY	.....GGEIDR	TACT	GGVTPHEGNLKERKK
SEE	...SDQF	ENTLLFKGFFTG	HPWYNDLLV	LGSKDATNKYK	GKKVDLYGAY	GYQCA	.....GGTPNK	TACM	GGVTLHDNNRLTEKK
SEB	...SIDQF	YFDLIYSIKDT	KLGNYNVRV	FKNKDLADKYK	DKYVDVFGAN	YYQCYF	SKKITNDIN	SHQTDK	GGVTEHNGNOLD..KY
SEC1	...SVDKF	AHDLIYNI	SDKKLKNYDKVKT	LLNEGLAKKYK	DEVVDVYGSN	YVNCYF	SSKDNVGV	KTG	GGITKHEGNHFDNGNL
SEC2	...SVDKF	AHDLIYNI	SDKKLKNYDKVKT	LLNEGLAKKYK	DEVVDVYGSN	YVNCYF	SSKDNVGV	KTG	GGITKHEGNHFDNGNL
SEC3	...SVDKF	AHDLIYNI	SDKKLKNYDKVKT	LLNEGLAKKYK	DEVVDVYGSN	YVNCYF	SSKDNVGV	KTG	GGITKHEGNHFDNGNL
SPEa	...SVDQF	SHDLIYNVSG	...PNYDKLKT	LLNQEMATL	FKKDNVGIYGE	YHLCYLC	ENAE	.....RSACI	GGVTNHEGNHLEIPK
TSST1	...VLDNS	GSMRIKNTD	.....GSISLI	FPSPYSPAF	TGKVDLNTKR	KKSQHT	SEB	.....TYIHF	QSGVTNT EKLPT...P

Fig. 3

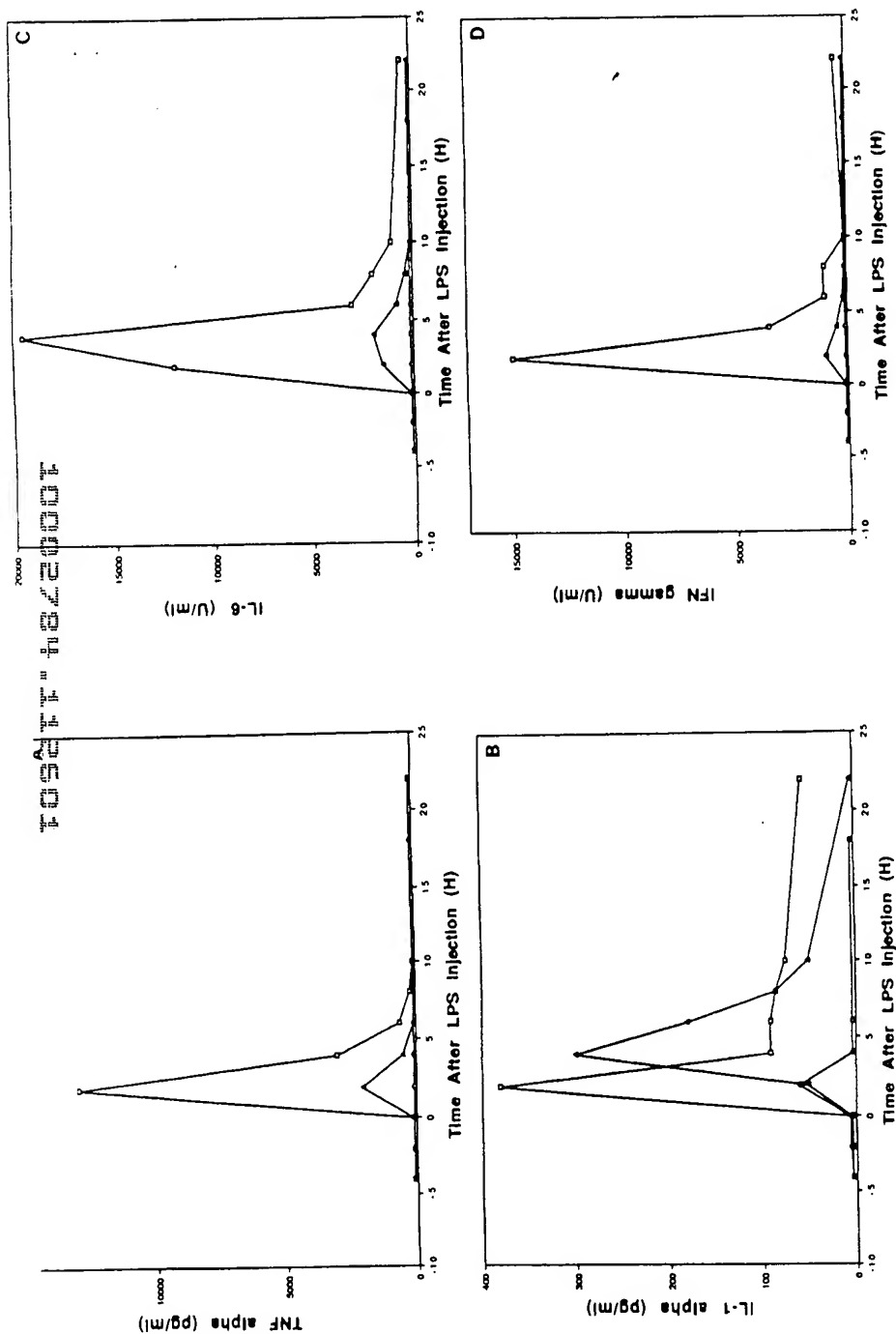


Fig 4

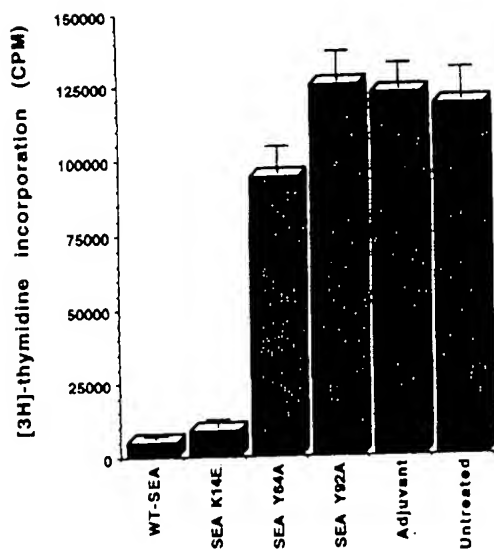


Fig. 5

TSST-1: 1622000

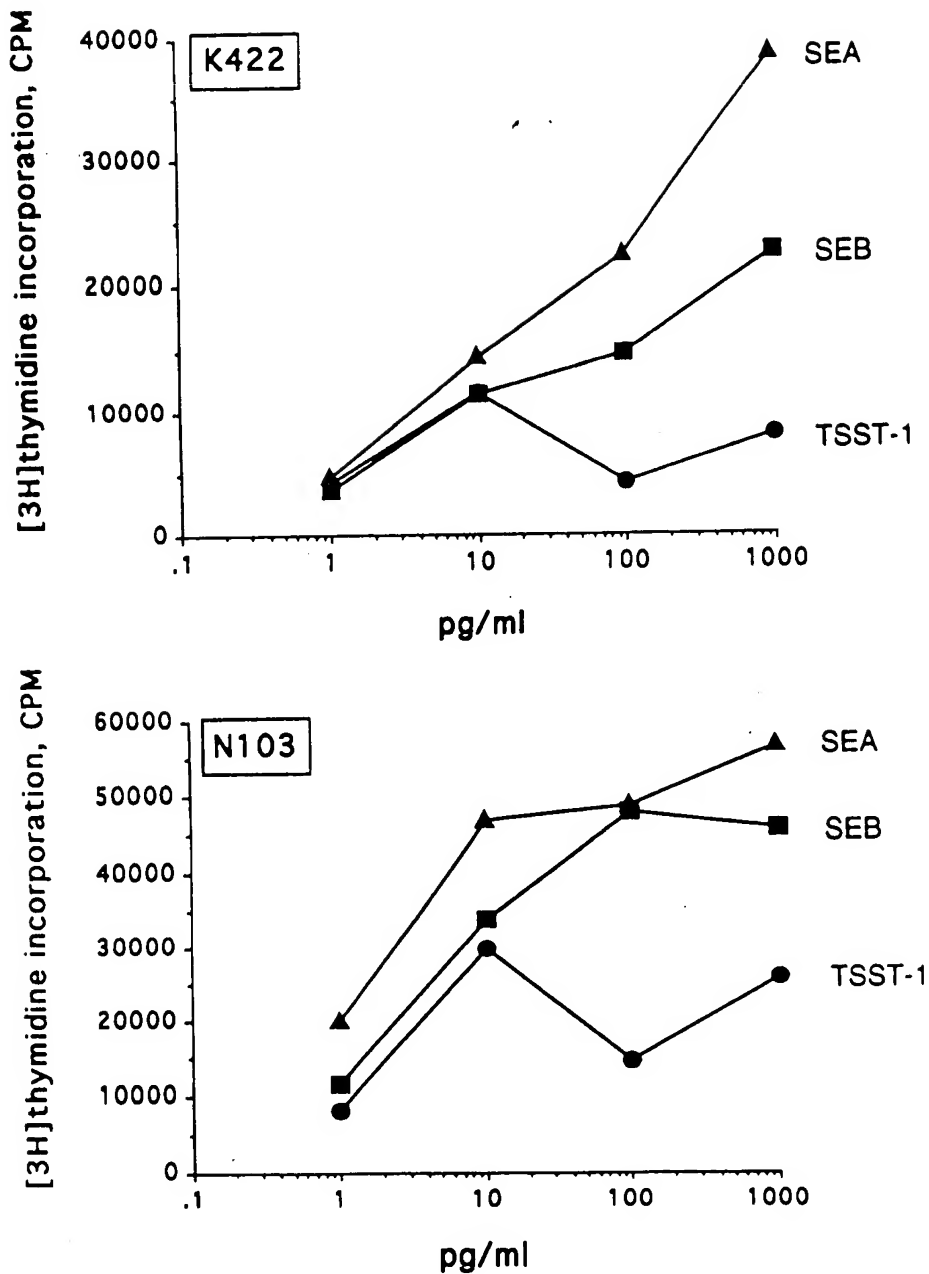
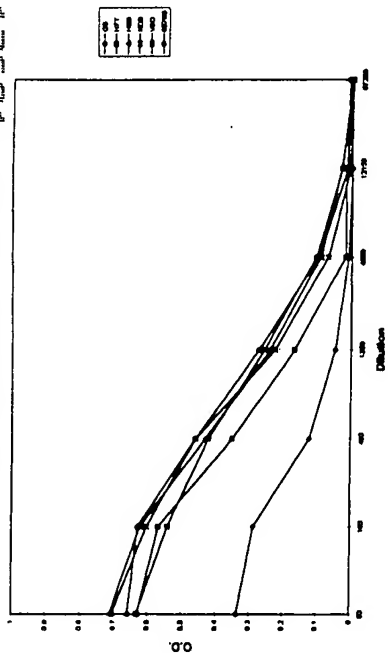
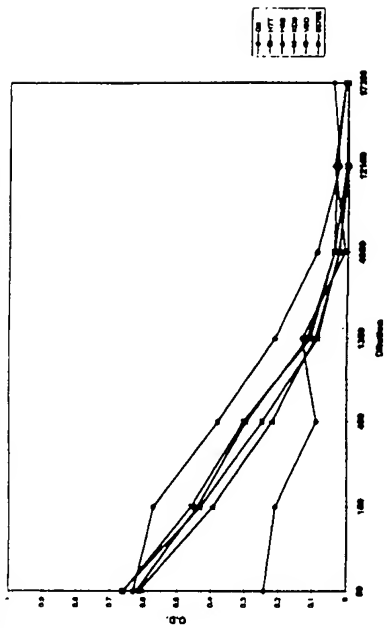


Fig. 6

Monkey Sera (3rd bleed) Response to SEA [0.25g/week]



Monkey Sera (3rd bleed) Response to SEC [0.25g/week]



Monkey Sera (3rd bleed) Response to SEC [0.25g/week]

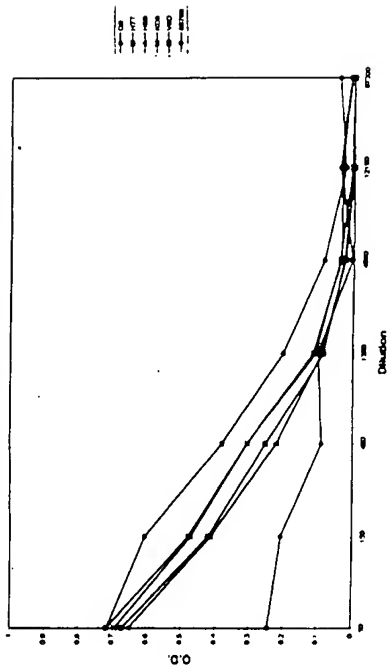
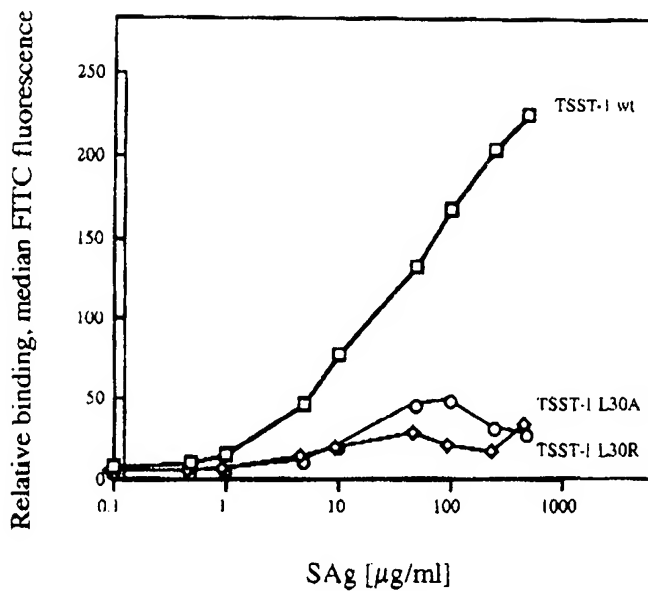


Fig 7

A.



B.

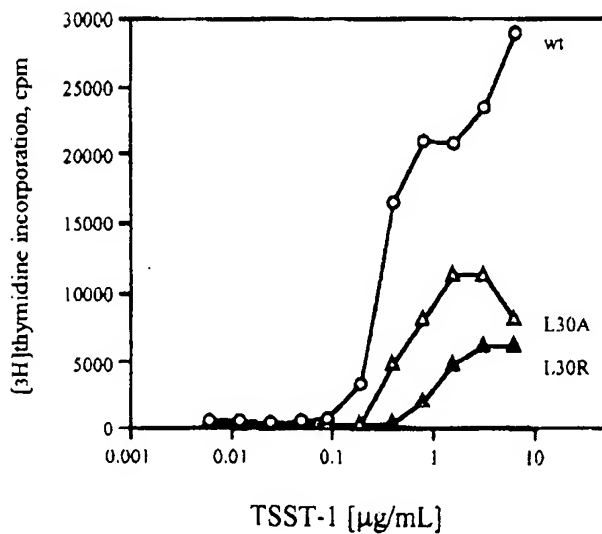
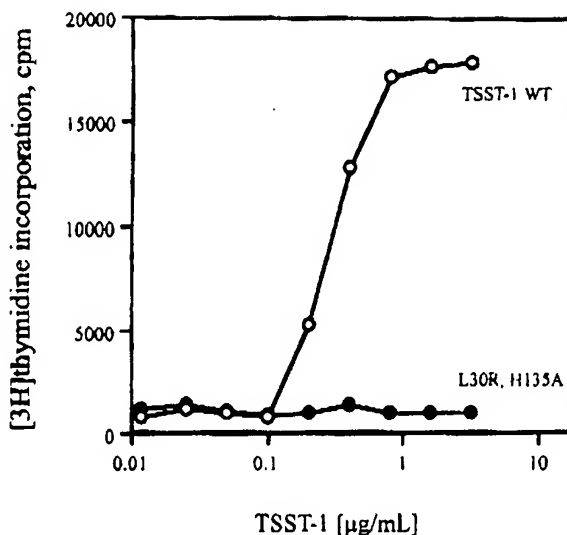


FIGURE 8

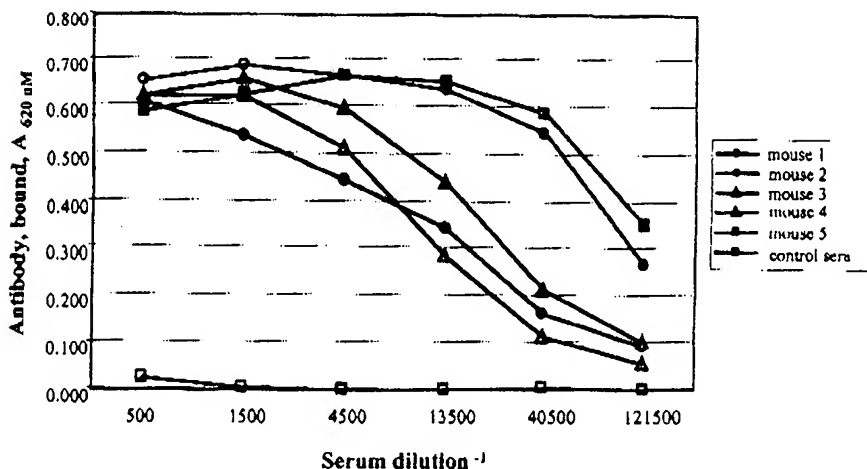


C.



**Biological activities of TSST-1 mutants.** a, Mutations of TSST-1 at amino acid position 30 (L30R, L30A) results in greatly diminished interactions with cell surface HLA-DR, measured by laser fluorescence-activated flow cytometry and FITC-labeled rabbit anti-TSST-1 antibody (affinity purified). b, Mutations of TSST-1 at amino acid position 30 (L30R, L30A) results in greatly diminished activation of human lymphocytes; c, Introduction of an additional mutation, H135A to the TSST-1 mutant L30R results in the maximum reduction in T-cell stimulation. Human T-cell proliferation, was assessed by [3H]thymidine incorporation, using a 12 h pulse with label and harvesting cells after 60 h of culture. Each data point represents the mean of triplicate determinations; SEM <5%.

FIGURE 8



**Antibody response to TSST-1 mutant L30R.** Mice received a total of three injections of vaccine (20 µg/mouse) in Alhydrogel, two weeks between injections. Sera were sampled two weeks after last vaccination and anti-TSST-1 specific antibody was measured by ELISA, using plates coated with wild-type TSST-1. Pooled non-immune mouse sera were used as negative control.

FIGURE 9

A.

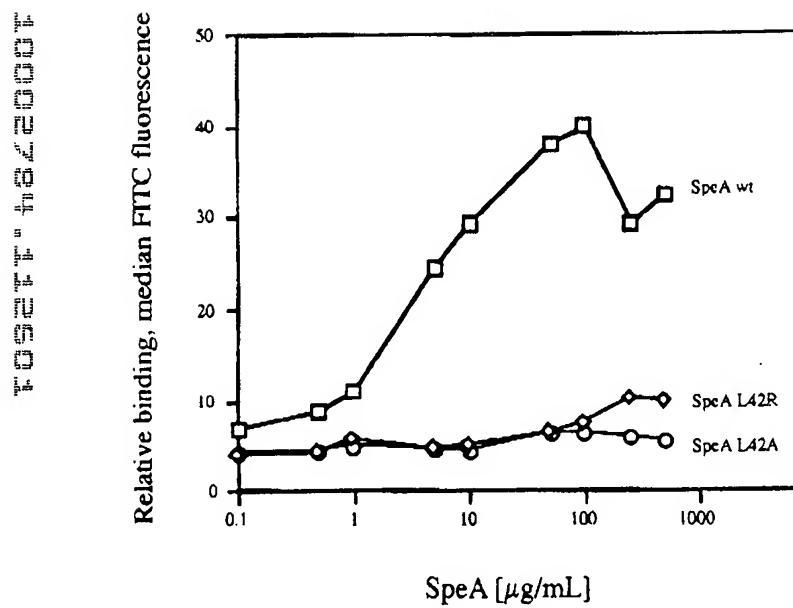
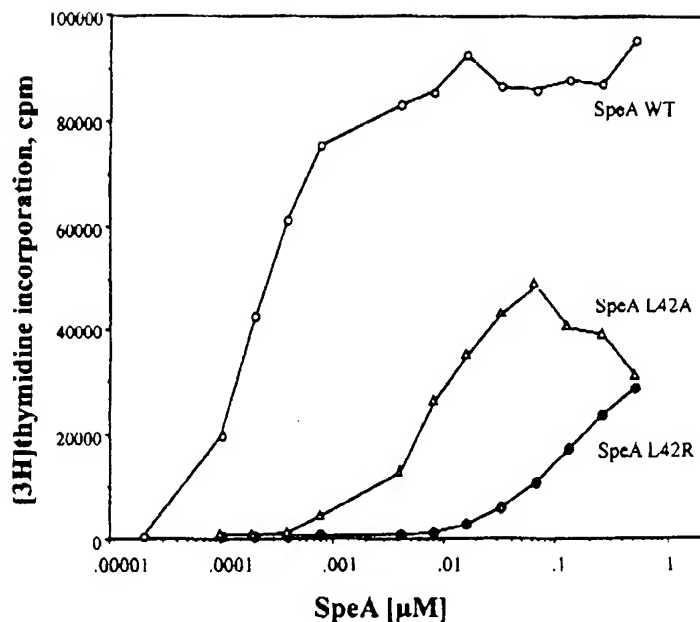


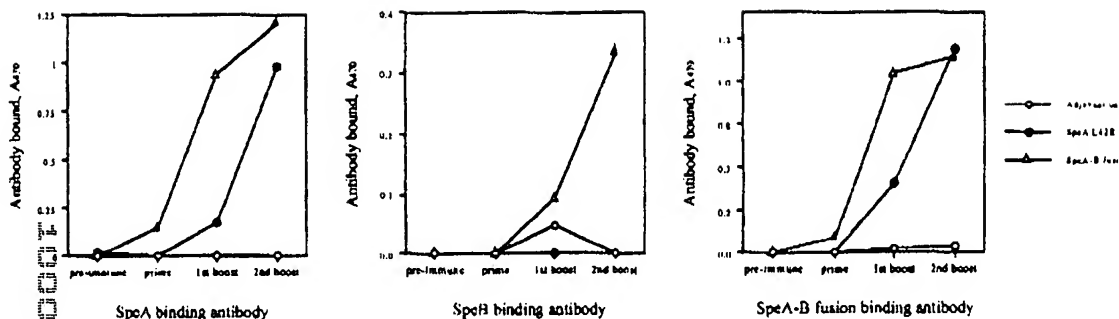
FIGURE 10

B.



**Biological activities of SpeA mutants.** a, Mutations of SpeA at amino acid position 42 (L42R) results in greatly diminished interactions with cell surface HLA-DR, measured by laser fluorescence-activated flow cytometry and FITC-labeled rabbit anti-SpeA antibody (affinity purified). b Mutations of SpeA at amino acid position 42 (L42R or L42A) results in greatly diminished activation of human lymphocytes. Human T-cell proliferation, was assessed by [3H]thymidine incorporation, using a 12 h pulse with label and harvesting cells after 60 h of culture. Each data point represents the mean of triplicate determinations; SEM <5%.

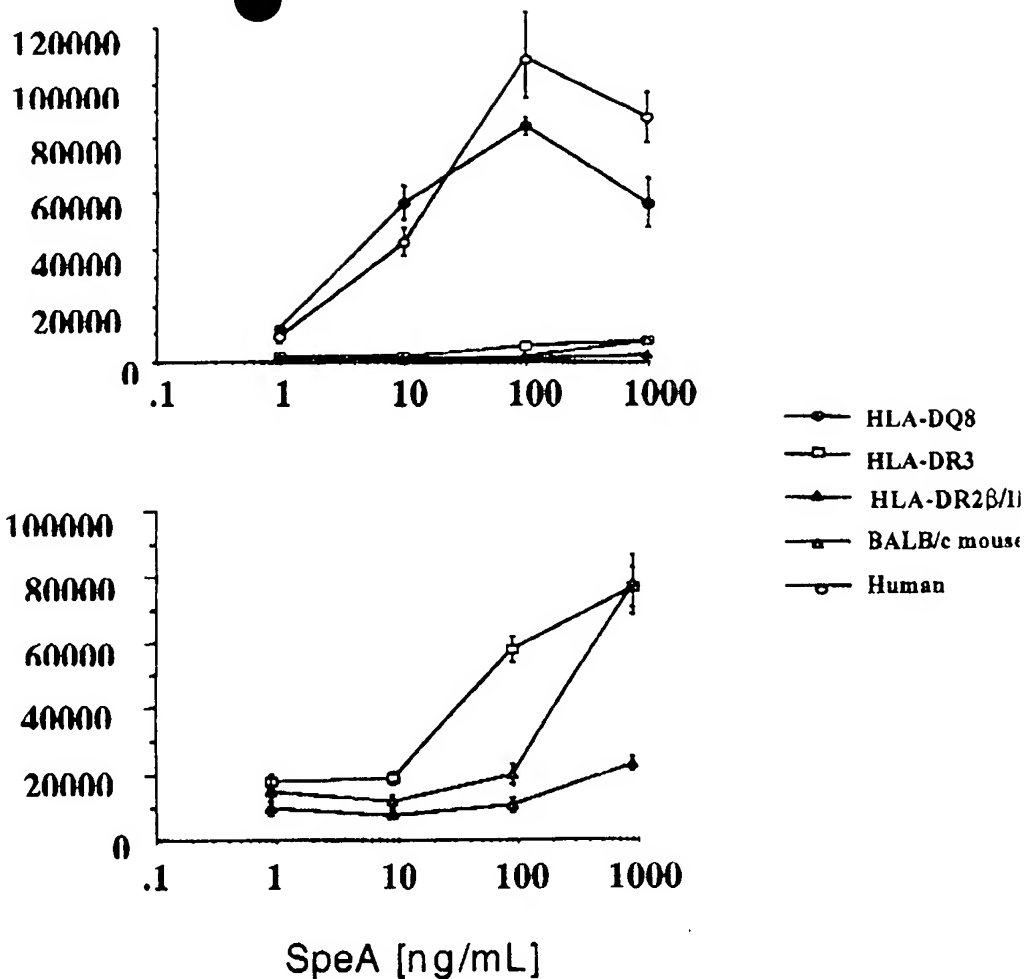
FIGURE 10



**Mouse antibody response to SpeA L42R and SpeA-B fusion constructs.** BALB/c mice were vaccinated three times with 10  $\mu$ g plus adjuvant (MPL™ + TDM+ CWS Emulsion, RIBI ImmunoChem Research, Inc., Hamilton, MT) of each construct, allowing two weeks between injections. Sera from each experimental group (n=5) were pooled for measurement of specific antibodies. Data shown are antigen-specific antibodies (ELISA units) present in a 1:100,000 dilution of pooled sera from mice vaccinated with SpeA L42R, SpeA-B fusion or adjuvant only.

FIGURE 11

Total [3H]thymidine incorporation, cpm



T-cell response *in vitro* of mononuclear cells from transgenic mice expressing HLA-DQ8αβ and human CD4 closely approximate the physiological response of humans. Mononuclear cells were isolated from spleens of transgenic mice expressing HLA-DR3, HLA-DQ8 or HLA-DR2β/1Eα, or non-transgenic BALB/c mice and human peripheral blood ( $4 \times 10^5$ /well). Following 60 h culture with SpeA, cells were pulse-labeled (12 h) with 1 μCi of [3H]thymidine. DNA from cells was harvested onto fiberglass filters and incorporated radioactivity measured by liquid scintillation.

FIGURE 12